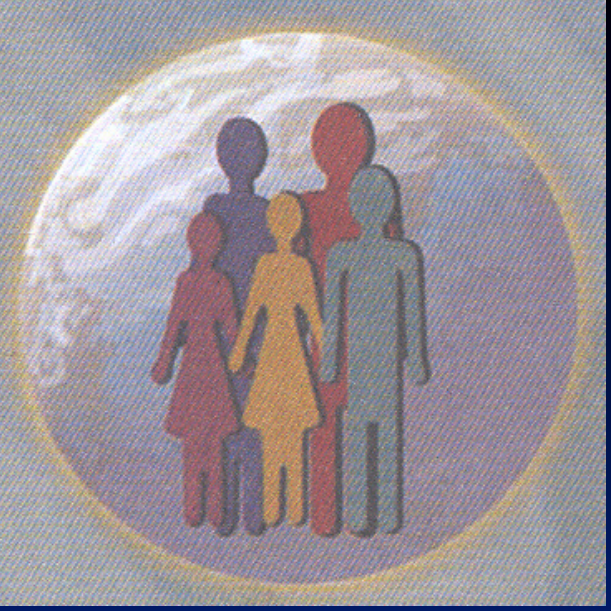


COLON CFR

PURPOSE:

Build an infrastructure to facilitate multi-disciplinary, collaborative research on the causes, prevention, and treatment of colorectal cancer

CENTERS

Australia Consortium
PI: John Hopper, Jeremy Jass

Cancer Care Ontario, Canada
PI: Steve Gallinger

Fred Hutchinson Cancer Research Center, Seattle
PI: John Potter

Mayo Clinic
PI: Laney Lindor

Cancer Research Center of Hawaii
PI: Loic LeMarchand

USC Consortium
PI: Robert Haile
Co-PI: John Baron

CORE DATA ITEMS

- Pedigree
- Blood sample (including ficoll-separated lymphocytes-cell lines)
- Risk factor questionnaire
- Food frequency Questionnaire (most centers)
- Tumor blocks and path reports for cases
- Fresh Frozen Tissue on 300 cases

COLON CFR DATA COLLECTION TO DATE

- # Population-based probands; 10,786
- # Clinic-based probands; 843
  - AmsterdamI = 508
  - Amsterdam2 = 635
  - Bethesda = 3,934
- #Risk factor questionnaires; 34,783
- # Blood samples collected; 20,120
- #Subjects with tissue blocks collected; 4,490

MAJOR RESEARCH AREAS

- Etiological Research
- Clinical Research (Molecular Profiles)
- Behavioral Research
- Prevention Trials

MSI (n = 5,417)

MSI-H MSI-L MSS

Studies of Risk Factors
(genes, environment, pathology)

Clinical Prognosis
And Response to
Treatment

MSI+

MSS

Germline Mutations in MMR Genes

Hypermethylation of MLH1 Promoter

dHPLC screening + seq. + GMP/MLPA
Methylight
- all clinic-based probands
- all population-based MSI-H, MSI-L
- 100 population-based MSS
n = 1900 probands

dHPLC + seq. + GMP/MLPA
Methylight

Mutation Carriers

MLH1 Hypermethylated

"normal" sequence

Combination of Selected Variables

Family History +

MSI +

IHC +

MMR Mutation Data +

hMLH1 Methylation

On = 2,000 Families

Carrier Prediction:

P (MMR Mutation | Family History)

P (MMR Mutation | MSI)

Sensitivity 96% - 100%

Specificity 60% - 22%

P (MMR Mutation | IHC)

Sensitivity 90% - 100%

Specificity 85% - 98%

Translational Significance:

Algorithms to select subjects for genetic tests

HNPCC

- Defined by Family History
- Defined by MMR defect

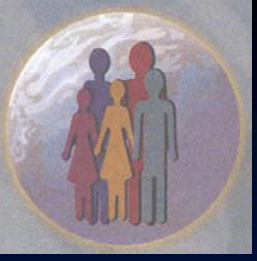
HNPCC

With MMR Defect

Cancer Risks?

No MMR Defect

Cancer Risks?



DHPLC + seq. + GMP/MLPA
Methylight

Mutation Carriers

MLH1 **Hypermethylated**

"normal sequence"

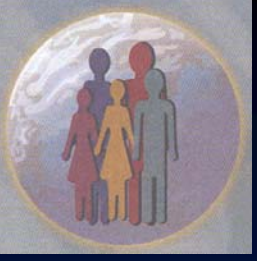
Gene characterization

- penetrance
- GXE
- GXG

- Environmental covariates
- other genes

CIMP

New Gene Mapping



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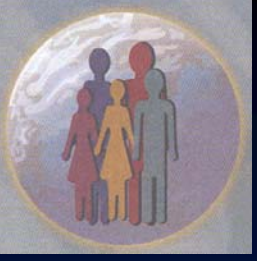

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- other genes

CIMP

New Gene Mapping

Gene Mapping

- Sib pairs (genome-wide scan)
- Finer mapping/positional cloning
- Sib pairs – TGFB (with Dr. Boris Pasche)

Gene Mapping

- Comparative genome hybridization
  - conventional CGH
  - germline CGH
- Proteomics
- Fresh Frozen and Paraffin-Embedded Tissue

Candidate Genes

- Focus on pathways not simple polymorphisms in a single gene
  - NSAIDs
  - Folate, vitamin D, calcium
  - Obesity, metabolic syndrome, IGF
  - Lipid peroxidation
  - HCAs, PAHs, nitrosamines

GENOME-WIDE ASSOCIATION STUDIES

(Complement candidate gene studies)

1. **ARCTIC**

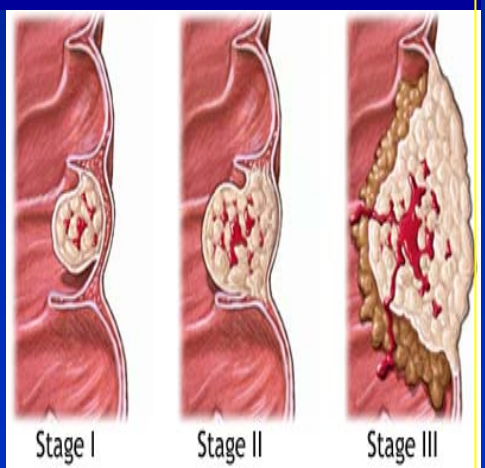
2. **CFR-Wide GWAS**

2005 Design:

Ontario CFR
•1200 Cases
•1200 Controls
• 1.4 billion genotypes

Validation Phase

•1200 Cases
•1200 Controls
• ~1500 SNPs



Output

•Predictors of Disease
•Disease prevention
•ARCTIC kits

Colon CFR-wide Genome-wide Association Study (GWAS)

- Multiple stages (take advantage of family-based CFR resource)
- Environmental/lifestyle data
  - probably relevant for common variant/common disease model
  - when and how to incorporate into analyses?

Epigenetic Epidemiology

- LOI of IGF2
  - very prevalent, especially in MSI-H cases
  - detectable in blood

  Goals:
  - Confirm prevalence
  - Estimate RR
  - Assess familial aggregation of LOI

- DNA methylation
  - MLH1 (2,469 samples completed)
  - CIMP (4,943 samples pending)

Behavioral Research
(examples)

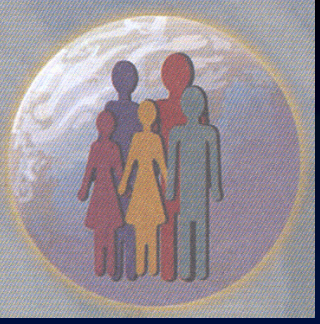
- Increase compliance with screening guidelines
- Patterns of knowledge and communication within families (heterogeneity by SES, ethnicity, health plans)

Clinical Research

Theme: molecular profiles …

1) MSI status and prognosis

2) Specific genes linked to specific treatments (e.g. TS and 5FU)



PENDING

MSI

TORONTO

Planned R01 (P.I. Neli Ulrich)

The specific aims of this proposal are:

1) To evaluate whether prognosis after stage I-III colorectal cancer (survival and recurrence) is related to folate status/DNA repair capacity of the cancer patient and the tumor.

   a. Folate intake
   b. Gene expression of Folate-metabolizing enzymes or DNA repair enzymes
   c. Polymorphisms in folate-metabolizing genes or DNA repair genes

2) To evaluate whether treatment outcomes after 5-FU (colon cancer patients stages II & III) or radiation therapy (rectal cancer patients stages II & III differ by

   a. Folate intake
   b. Polymorphisms in folate-metabolizing enzymes or DNA repair enzymes
   c. Gene expression of folate-metabolizing enzymes
   d. MSI or CIMP status

Possible CFR Prevention Trial

Need Collaborators for HNPCC Focus

- CFR
- INSIGHT
- CGA
- CAPP Consortium
- ?CGN
- ?Other centers (Creighton, etc)

Possible CFR Prevention Trial

Possible Agents

- Aspirin
- Other NSAID
- Calcium (± Vitamin D)
- Statin

Minority Populations

Epidemiologic Research on Ethnic/Racial Minorities in the Colon CFR (P.I. L. LeMarchand)

Focus: African Americans and Japanese Americans

Motivation:

- Japanese Americans and African Americans have a substantially elevated risk of CRC
- African-Americans have a shorter, stage-adjusted survival than white cases

Goals:

1, 514 African American families

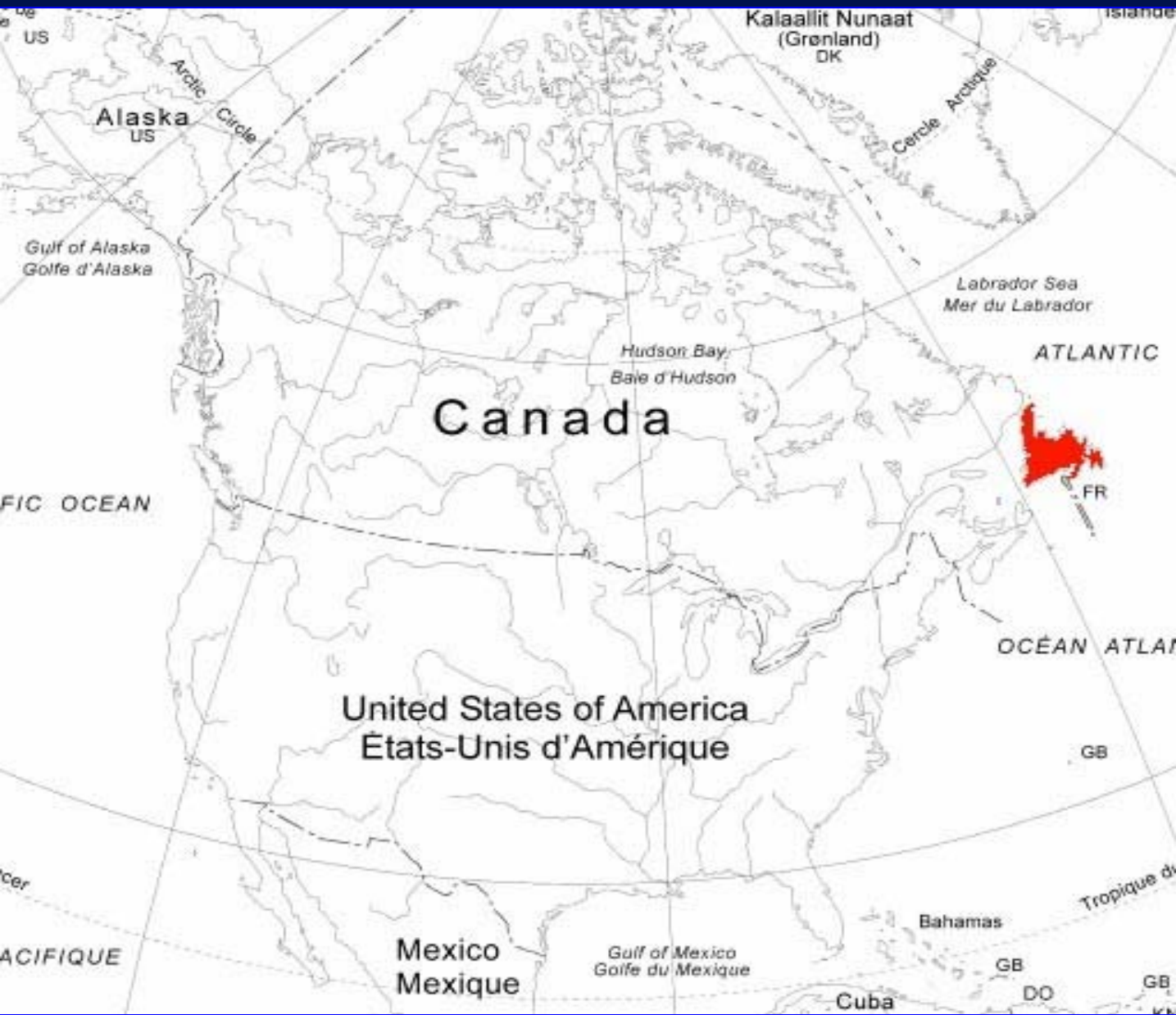
745 Japanese American families

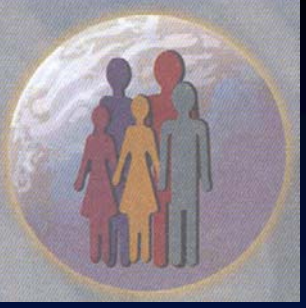
Core Data:

Environmental/lifestyle variables in Colon CFR

Extensive genotyping (see poster)

Location of Newfoundland





Newfoundland Population

- Population ~ 500,000
- Large families (until recently)
- Genetically isolated populations have proven to be a useful resource in discovering disease genes
- The Newfoundland Population is one of the very few North American populations that falls into this category
- The availability of good archival and medical records and the receptive and cooperative nature of the people have also contributed to numerous important human genetic discoveries
- 739 probands already enrolled

Reasons to consider the conduct of genetic-epidemiology studies of the Galician population

Advantages of the Galician Population for Conducting Genetic Epidemiological Studies: Relevance to Planned Initiatives

1. Genetic homogeneity of the Galician population

2. Galician population's migration history to Latin America

3. Galician organizations existing in several countries in Latin America where Galician communities can be identified

4. Excellent infrastructure in place to support studies

Infrastructure

1. The relatively large families in the generations we will study

2. Relatives of probands often live in the same household or town, and almost always in the same region – very cooperative

3. A centralized and universal health care system that should result in nearly complete case finding and virtually 100% retrieval of medical records
   -- Unique health ID# for each inhabitant of Galicia
   -- Very supportive clinicians/scientists

Future Plan
Cohort Study of MMR Mutation Carriers

1. Expand existing families
2. Recruit more families from within the Colon CFR
3. Collaborate with other groups

   e.g. **The UK CAPP2 (Colorectal Adenoma/Carcinoma Prevention Programme) Cohort**

   CAPP2 Details:

   Principal Investigator: Professor John Burn, University of Newcastle, UK

   Co-Investigators include:

   Tim Bishop (Leeds, UK; Role: Statistics)

   John Mathers (Newcastle, UK; Role: Nutrition, Study Design

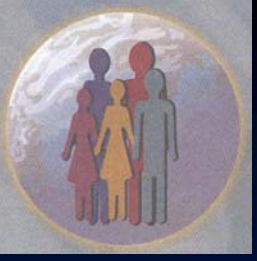
   n = 1000 mutation carriers

Research Potential To Be Realized

Early Detection

X = Incident Colorectal Cancer

Colon CFR = Higher Yield Cohort



Summary of Colon CFR Studies
70 approved, ongoing studies

GRANTS

NCI

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Creation of a Permanent Genetics Resource at the CFRCCS (P.I. J. Potter)

Epidemiologic Research on Ethnic/Racial Minorities in the Colon CFR (P.I. L. LeMarchand)

Genetic Linkage in Colorectal Cancer Families (P.I. J. Potter, P.I. of Mayo subcontract Ellen Goode

NSAID and COX/PG Metabolism and Colorectal Cancer (P.I. J. Potter)

Folate, Calcium, and Vitamin D in Colorectal Cancer (P.I. R. Haile)

Obesity, Metabolic Syndrome & IGF (P.I. E. Martinez)

Australian

Germline Mutations in Mismatch Repair Gene: Prevalence, Risk of Cancer, and Environmental Modifiers of Risk

(P.I. M. Jenkins, Univ of Melbourne; L. Baglietto, Cancer Council Victoria; T. Bishop, Cancer Research Leeds; I. Winship, Genetic Health Services, Victoria; M. Barker, Queensland Inst of Med Research)

Canadian

ARCTIC (P.I. Brent Zanke and Tom Hudson)

The MYH Gene and Colorectal Cancer Risk

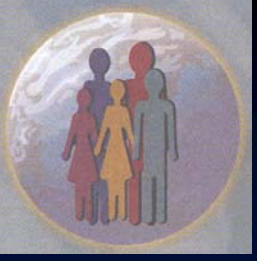
(P.I. S. Gallinger, National Cancer Institute of Canada)

PENDING

Lipid Peroxidation (M. Gago)

GWAS (G. Casey)

Meat Intake, Genetic susceptibility, & colorectal cancer risk (M. Stern)



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PostDocs (partial list)

Jenny Poynter – USC

Charlotte Onland-Moret – USC

Margreet Luchtenborg – Cancer Research Center of Hawaii

Shinichi Hayashi – McGill

Virginia Kaklamani – Northwestern

Yanfei Xu – Northwestern

Sarah Harrington – University Health Network & Mt. Sinai

Michelle McGreevy – Cleveland Clinic

Libby Morimoto – Fred Hutchinson Cancer Research Center

Sir Paul Doria Rose – Fred Hutchinson Cancer Research Center

Brenda Diergaarde – Fred Hutchinson Cancer Research Center

Rebecca Sedjo – University of Colorado

Lucia Fini – Baylor

Xiaofei Shi – University of Toronto

Rob Gryfe – Mt. Sinai Hospital, Toronto

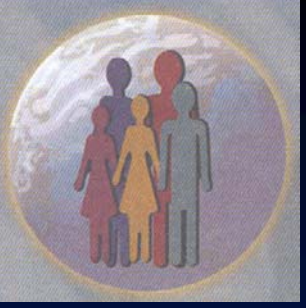
George Zogopoulos – Mt. Sinai Hospital, Toronto

Pam Sinicrope – Mayo Clinic

Csilla Szabo – Mayo Clinic

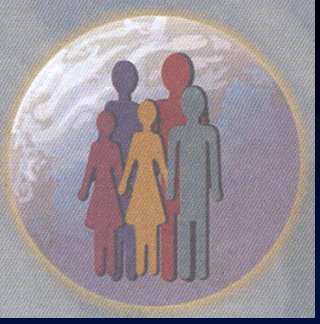
Collaborative Studies (Beyond Original CFR Centers)

P.I.	Institution	Title
Boland	Baylor University	Using CFR Resources to Study HNPCC
Feinberg Haile	Johns Hopkins	A Molecular Epidemiology Study of Loss of Imprinting
Frazier	U. of Texas	Genetic Modifiers of Hereditary Nonpolyposis Colorectal Cancer
Kohut	Sarah Lawrence/ Mt Sinai Hospital	Duty to Warn Family about an HNPCC Mutation
Madlensky	UCSD	Health Behaviors and Family History of Colorectal Cancer
Parmagiani	Johns Hopkins	Validation of the CRCAPRO Carrier Probability Model
Pasche	Northwestern	Polymorphisms of the TGF-β Signaling Pathway and Colorectal Cancer Risk
Scheuner	Ctr Dis Ctr & Prev	Clinical Validity Study of Colon Cancer Family History
Siciliano	U. of Texas	MSI in Putatively Stable HNPCC Families



You are invited to

Collaborate . . .



Collaborating is fun!